I. SUPPORT FOR THE AMENDMENT AND NEW CLAIM

Support for the amendments and new claim is found through the specification as originally filed. More particularly, support for the amendment to the claims is found, *inter alia*, on page 8, lines 23-32, bridging to page 9, lines 1-32. Support for new claim 59, is found, for example, in claims 32 and 35 as filed. No new matter has been introduced with the foregoing amendments and newly added claim As such, Applicants respectfully request that they be entered.

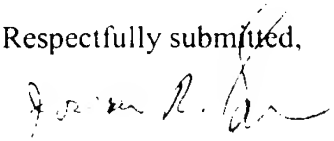
II. THE INVENTION

The present invention provides methods for large-scale sialylation of saccharide groups present on a glycoprotein. The methods comprise contacting the saccharide groups with a sialyltransferase, a sialic acid donor moiety, and other reactants required for sialyltransferase activity for a sufficient time and under appropriate conditions to transfer sialic acid from the sialic acid donor moiety to the saccharide group.

III. CONCLUSION

In view of the foregoing, Applicants respectfully request early action on the merits. If the Examiner believes a telephone conference would expedite prosecution of this application, please telephone the undersigned at 925-472-5000.

Respectfully submitted,



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VERSION WITH MARKINGS TO SHOW CHANGES MADE

In the Specification:

Please rewrite the first paragraph on page 1 as follows:

This application is a continuation of U.S. Patent Application No. 09/007,741, filed January 15, 1998, allowed, **[This]** which application claims priority to US Provisional Application 60/035,710, filed January 16, 1997, which is incorporated herein by reference in their **[its]** entirety for all purposes.

Please rewrite the paragraph at page 2, line 3 as follows:

Production of glycoproteins in transgenic animals has some of the same problems as mammalian cell culture. While the "production" of a glycoprotein is inherently better controlled, it is also less susceptible to manipulation. If glycosylation is not complete, there is little that can be done with the animals to alter the outcome. With transgenic animals there is often another problem. While the predominant sialic acid in humans is N-acetyl-neuraminic acid (NeuAc), goats, sheep and cows all produce a large fraction of their total sialic acid as N-glycolyl-neuraminic acid (NeuGc). Although the impact of this modification is not yet fully explored from a functional or regulatory perspective, it is known that the NeuGc substitution is antigenic in humans (Varki (1992) *Glycobiology* **2:** 25-40).

Please rewrite the paragraph at page 3, line 5 as follows:

Sialyltransferases that are useful in the methods of the invention typically have a sialyl motif that comprises about 48-50 amino acids, within which about 40% of the amino acids are identical to the consensus sequence RCAVVSSAG---DVGSKT (where --- indicates a variable number of amino acid residues such that the motif is about 48-50 residues in length). Examples of sialyltransferases that are suitable for use in the present invention include ST3Gal III (preferably a rat ST3Gal III), ST3Gal IV, ST3Gal I, ST6Gal I, ST3Gal V, ST6Gal II, ST6GalNAc I, ST6GalNAc II, and ST6GalNAc III (the sialyltransferase **[nomemclature]** nomenclature used herein is as described in Tsuji *et al.* (1996) *Glycobiology* 6: v-xiv). The methods of the invention can involve sialylation of recombinant glycoproteins with more than

one sialyltransferase; for example, with an ST3Gal III and an ST3Gal I, or an ST3 Gal III and an ST6 GalI, or other combinations of enzymes. The sialic acid donor moiety used in the claimed methods is generally CMP-sialic acid, which can be added to the reaction directly or can be enzymatically generated *in situ*. The sialic acids used in a preferred embodiment are selected from the group consisting of NeuAc and NeuGc.

Please rewrite the paragraph at page 3, line 5 as follows:

Ara	= arabinosyl;
Fru	= fructosyl;
Fuc	= fucosyl;
Gal	= galactosyl;
GalNAc	= **[N-acetylgalacto]** N-acetylgalactosaminyl;
Glc	= glucosyl;
GlcNAc	= **[N-acetylgluco]** N-acetylglucosaminyl;
Man	= mannosyl; and
NeuAc	= sialyl (typically N-acetylneuraminyl).

In the Claims:

Please amend claims 1, 23, 32, and 57 as follows:

1. (Amended) A commercial-scale production method of sialylating a saccharide group on a recombinant glycoprotein, the method comprising contacting a saccharide group which comprises a galactose or N-acetylgalactosamine acceptor moiety on a recombinant glycoprotein with a sialic acid donor moiety and a recombinant sialyltransferase in a reaction mixture which provides reactants required for sialyltransferase activity for a sufficient time and under appropriate conditions to transfer sialic acid from said sialic acid donor moiety to said saccharide group.

23. (Amended) A commercial-scale production method of sialylating a saccharide group on a recombinant glycoprotein, the method comprising contacting a

saccharide group which comprises a galactose or an N-acetylgalactosamine acceptor moiety on a recombinant glycoprotein with a sialic acid donor moiety and a bacterial sialyltransferase in a reaction mixture which provides reactants required for sialyltransferase activity for a sufficient time and under appropriate conditions to transfer sialic acid from said sialic acid donor moiety to said saccharide group.

32. A commercial-scale production method for *in vitro* sialylation of saccharide groups **[present]** on a glycoprotein, said method comprising contacting said saccharide groups with a sialyltransferase, a sialic acid donor moiety, and other reactants required for sialyltransferase activity for a sufficient time and under appropriate conditions to transfer sialic acid from said sialic acid donor moiety to said saccharide group **[, wherein said sialyltransferase is present at a concentration about 50 mU per mg of glycoprotein or less]**.

57. A commercial-scale production method for *in vitro* sialylation of saccharide groups **[present]** on a glycoprotein, the method comprising contacting the saccharide groups with an ST3Gal III sialyltransferase, a sialic acid donor moiety, and other reactants required for sialyltransferase activity for a sufficient time and under conditions to transfer sialic acid from said sialic acid donor moiety to said saccharide group **[, wherein said ST3Gal III sialyltransferase is present at a concentration of less than about 50 mU per mg of glycoprotein]** .

59. (New) A commercial-scale production method for sialylation of saccharide groups on a glycoprotein, said method comprising contacting said saccharide groups with a sialyltransferase, a sialic acid donor moiety, and other reactants required for sialyltransferase activity for a sufficient time and under appropriate conditions to transfer sialic acid from said sialic acid donor moiety to said saccharide group, and wherein at least about 80% of the saccharide groups are sialylated.